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Pyrethrin Type Compounds from the Municipal Solid Waste Ghazipur Landfill Inert Soil as Brain and Cancer 5-HT Receptor Binding Agents

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Abstract

The Inert soil obtained from the Ghazipur Municipal Solid Waste landfill located in East Delhi, India has been used as the starting material for the work described in this paper. This mountain-like monster is often in the news for its bad smell, pollution of the groundwater in the area from its leachates, and the occasional fires in it endangering the lives of the people living nearby. Landfills produce many greenhouse gases (CO_2, CH_4, CO, HCHO, H_2S, and NH_3) which are poisonous and cause environmental pollution in the surrounding areas. Our work sought alternative uses for this waste material ("Waste to Wealth"). These studies could lead to a cleaner environment, lower carbon footprints, diminished global warming, and positively impact climate change. These are very hot topics being discussed under the sustainable development goals and COP-28 urging, in particular, the developed world to reduce their carbon footprints and to create a global fund to mitigate the climate crisis in underdeveloped countries caused by climate change. Four new Pyrethrin-like compounds have been isolated and characterized spectroscopically [using UV-visible, FT-IR, NMR spectroscopy (^{1}H-NMR

spectroscopy), Mass Spectrometry, and Scanning Electron Microscopy (SEM)]. The new compounds possess a cyclopropane ring substituted on one side by a 4-methoxy-3-oxo-pyruvate unit and on the other side by an N-formyl-amino acid unit. The presence of pyrethrin-like cyclopropane structures indicates possible insecticidal activities like natural pyrethrins. The N-formyl amino acid could be an important factor in their bioactivity. Computational studies using the software Chem J. predict antibacterial, and anti-inflammatory activities with an IC_{50} value of 8.720.

Keywords
Humic substances; mass spectrometry; NMR spectroscopy; cyclopropane natural insecticides; computational studies

1. Introduction

Landfill sites in India and the world are increasing dramatically due to the production of different types of garbage locally, nationally, and internationally. Due to this, the Contemporary Landfill management system is continually under pressure and has raised an alarm among the international community. To confront this, many forums and conferences have been organized worldwide [1]. Issam A. Al-Khatib et al. describe the MSW conditions in the seven major northern West Bank districts and Palestinian territory [2]. Raquel Alfaia, G.D.S.M., et al. have discussed the panorama concerning the situation of MSW in Brazil from generation to final disposal. In addition, they have provided an in-depth analysis of the issues that caused delays in implementing of the National Policy of Solid Waste (NPSW) [3]. Kahvand, M. et al. discussed a suitable solution using a computer and GIS related to solid urban waste and their location in Hamadan based on the standard criteria [4]. Verma, R.L. et al. performed a SWOT (Strengths, Weakness, Opportunities, and Threats) analysis of MSW in Ho-Chi-Minh City, Vietnam [5]. Vergara, S.E. et al. have discussed the present state of affairs regarding MSW management globally [6].

The landfills mostly available today employ traditional dumping methods and produce many dangerous greenhouse global warming gases like Carbon dioxide (CO_2), Methane (CH_4), Carbon monoxide (CO), HCHO (a known carcinogen), Ammonia (NH_3), and Hydrogen Sulfide (H_2S), along with highly hazardous products, such as leachates coming out of the landfill [7] (Figure 1).

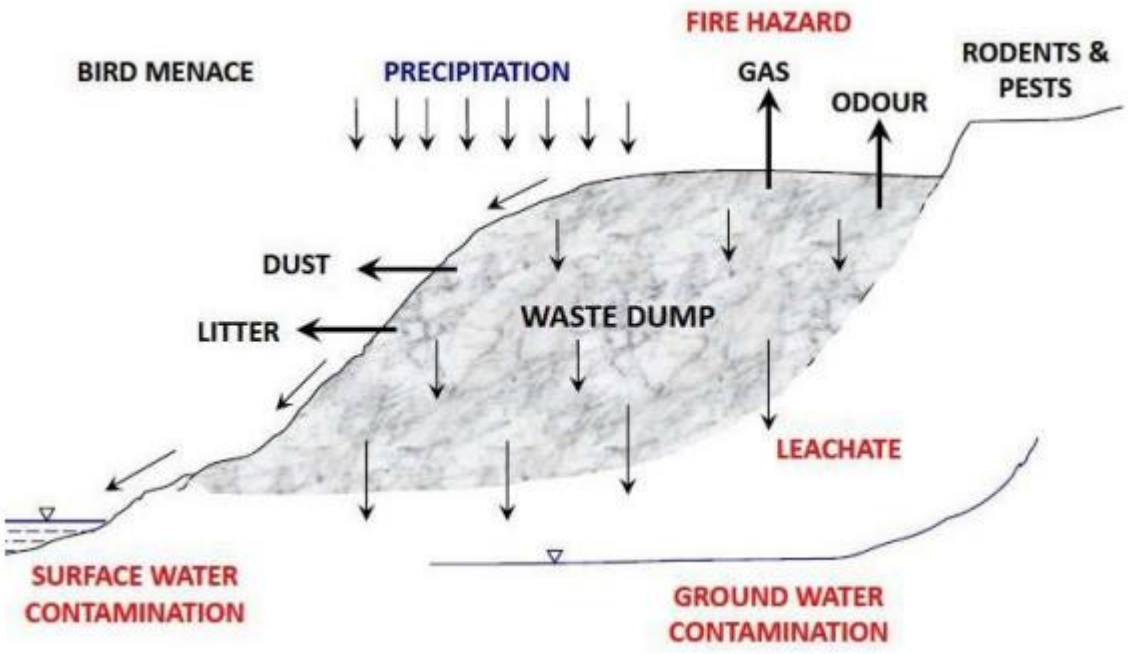

Figure 1 Schematic of old landfilling.

To overcome these problems associated with old landfilling sites many countries have begun to make a transition to modern landfilling sites with an improved infrastructure, which are developed to minimize the environmental impacts and safeguard public health and locality (Figure 2).

Figure 2 Schematic of Modern landfilling.

Modern landfilling consists of a series of lift bents of 50 ft. to 75 ft. including the cover layer which can be closed after the operations are over. The work is done in five phases described as initial adjustments consisting of the Gas vent pipe, Topliner system, Surface vegetation, Collector pipe, Anchor trench, Side liner system, Leachate collection and removal system, and bottom line system [8].

Landfills hamper ecology due to the emission of poisonous gases, and foul smells and cause water pollution in the nearby area. This situation has led to environmental, social, and significant health issues which are present globally. Leachate has been defined as "formed when rainwater filters through wastes placed in a landfill. When this liquid comes in contact with buried wastes, it leaches, or draws out, chemicals or constituents from those wastes". Occasional fires are caused spontaneously or otherwise endanger the lives of nearby people [9]. On occasions, lives have even been lost. Many Municipal Corporations issue advisories to people living near the landfills, including the advice that they should not open windows towards the landfill site. In addition, they are advised to fill all cracks in the house walls and cement any cracks on the floor, to prevent the noxious gases from entering the house [10]. This issue dominates many conferences, global meets, and even the United Nations meetings. Listed below (Table 1) are the many pollutants emitted during dumping, incineration, composting, recycling, and transportation.

Table 1 Environmental impacts of various waste technologies.

Environmental Sink	Dumping/landfill	Incineration	Composting	Land application	Recycling	Transport
Air	CO, CH_4, odour, Noise, VOCs, GHGs (CO_2, CH_4, N_2O)	SO_2, N_2O, HCl, CO, CO_2, Dioxins, Furans, PAHs, VOCs, GHGs, Hg	Odour, GHGs (minor)	Bio aerosols, Odour, GHGs (minor)	GHGs (minor)	CO_2, SO_2, NO_x, odour
Soil	Heavy metals, organic compounds	Fly ash, Slag	Minor impact	Bacteria, Viruses, Heavy metals, PAH, PCBs	Landfilling of residues	
Water	Leachate, heavy, metals, organic compounds	Fallout of atmospheric pollutants	Leachate	Bacteria, viruses, heavy metals	Waste water from processing	Fallout of atmospheric pollutants (e.g., Nitrate)

Among the suggested solutions to the problem is to decimate the landfills. But the problem will remain with where and how to dump the MSW, whose amounts are increasing yearly. An alternative strategy is the Government of India's "Waste to Wealth" program, which aims at generating wealth from such waste. Plants have been set up which generate electricity using this waste. Another

approach is to convert these materials into new types of strengthened bricks for laying roads [11]. The magnitude and size of this problem can be gauged from the fact that each landfill produces millions of tons of such waste and each city in India has multiple such landfill sites. One can imagine the size of this source in each country and globally. Most publications in this study area focus mainly on describing the problem, particularly the socio-economic aspects. The print news and social media sensationalize the issue with obvious political overtones. Some other innovative approaches currently employed to solve this problem include, using this waste material to produce cheap electricity, In India Bricks are being formed to lay roads and buildings, and another approach is to decimate these mountain-like structures around the major cities [12-15]. However, are these solutions enough? The City solid waste will continue to be generated in large quantities, even if, some technologically useful products are prepared from the landfill-inert soil, these will be coming substantially from the residual material. It is thus clear one still has to look for a simpler method of reducing foul smell, stench, and leachates from such a source. The method used must also quell the generation of these toxic gases. An alternative possibility to be considered is whether these gases can be trapped and neutralized in a tank reactor attached to a landfill. This could also apply to leachates which could be collected and then neutralized. The concept of the use of a covered landfill treatment plant is discussed below:

There are clear EPA's RCRA laws and regulations that govern such landfills. Such landfills should not be made on flood plains, wetlands, etc. [*loc. cit*]. It should consist of a flexible membrane overlaying 2 feet of compacted clay soil at the landfill's bottom to protect groundwater from leachates. It is suggested that leachates be collected and treated before disposal, which will help reduce foul smell and the number of insects and rodents. Groundwater in the area must be tested from time to time. The Governmental agencies must ensure enough budget allotments for building such closed landfills. There must be strict regulations regarding the transport of MSWs, it is even suggested that these could be collected and re-transported from intermediate stations.

Unlike the normal landfills which are open to the skies, the covered landfill will have a cover like a roof. It is suggested that, unlike the mountain-like structures seen presently, these could be smaller thousand-liter reactors made of a suitable material (steel/ glass/ fire retardant plastic). In addition to a stirrer, the reactor must have a heating arrangement (50-60°C). There must be an outlet from which leachates can be collected for further treatment. This presumes that on the top of the cover, there must be a connection for water to be filled in, such that it can be removed as a leachate. The Plant should have an attached automatic methane measurement gadget to ensure that the spontaneously inflammable methane gas does not cross the required limit to prevent fire accidents. On top of the plant, there must be an outlet for gases to be collected as fuels for further distribution. After optimization, such a plant could be set up in every residential locality, ensuring that household green wastes are collected in such a plant within the apartment (Figure 3).

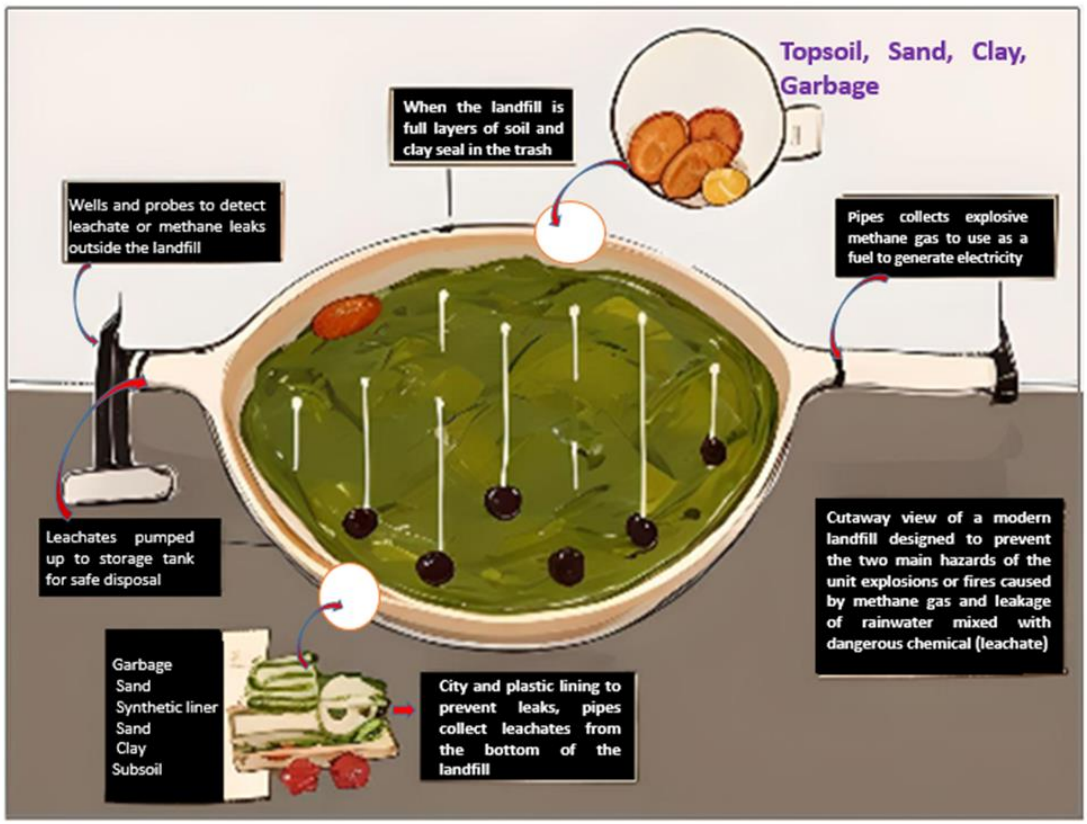

Figure 3 A diagram of the closed modern landfill site.

The Residential Welfare Association of East Delhi has taken a similar step in collaboration with MCD and the Indian Pollution Control Association (IPCA) for mitigating the waste collection in residential areas. There are approximately 100 societies in East Delhi that are involved in the segregation of waste sources. These reactors produce compost from biodegradable waste via the installation of aerobic composters. One aerobic composter with a capacity of 500 liters is provided to every 35 households in a society [16], which is a good beginning.

Our present research work:

This research paper aims to find out the major constituents present in one such landfill and whether any of these materials could be produced as value-added products for use in pharmaceuticals, biomedicine, agriculture, and materials science. Another aim is to neutralize key components before their breakdown in the landfill, which could harm the population. We studied the inert soil from the Ghazipur Municipal Solid Waste landfill in East Delhi. The material was provided to us by the East Delhi Municipal Corporation (EDMC). This mountain-like monster (Figure 4) produces a foul smell that spreads over many kilometers.

Figure 4 A picture of the Ghazipur landfill.

This landfill is often in the news for the wrong reasons. As of 2022, it has already touched the height of the Taj Mahal and the Qutab Minar and lives have been lost in occasional fires at the site [17].

For isolating these compounds, we followed the protocol of the International Humic Substances Society (IHSS) with some modifications where necessary. The products have been characterized using modern spectroscopic techniques described in the paper [18-20]. The chemical structures are most unusual and not described earlier in the literature. The variation among these structures indicates the strong possibility that carbon monoxide (a poisonous gas that affects respiration) and formaldehyde (a known carcinogen) could be generated during the degradation of these key components identified in this landfill-inert soil. Value-added products could be prepared that could serve as insect repellants, and anti-bacterial agents, as some of these compounds possess pyrethrin-like structures, which have led to environmentally safe insecticides (Allethrin, Cypermethrin, and Deltamethrin).

Computational studies point towards anti-cancer activity and the ability of these compounds to cure brain diseases (Parkinson's, Dementia, and Alzheimer's).

2. Materials and Methods

All the reagents and solvents were purchased from Sigma Aldrich (greater than 99.9% purity). The UV-visible spectrum was recorded in methanol at room temperature using a Perkin Elmer Lambda 35 double-beam spectrophotometer. SEM was also recorded using the SEM Zeiss EVO MA 10 in a methanol solution. Initial ^{1}H-NMR studies were done in the International Centre For Genetic Engineering and Biotechnology (ICGEB, New Delhi) & the Translational Health Science and Technology Institute, Faridabad (THSTI, Faridabad).^{1}H-NMR, was recorded using Bruker 500 MHz NMR instrument in D_2O solution in Tata Institute of Fundamental Research (TIFR), Hyderabad. The mass spectrum of this compound was recorded in THSTI, Faridabad, and the MALDI-MS spectrum

was recorded in CSIR–Indian Institute of Chemical Technology (CSIR-IICT), Uppal Road, IICT Colony, Tarnaka, Hyderabad, Telangana 500007. FT-IR studies were performed using Bruker Tensor 27 FT-IR with Diamond ATR cell in methanol solution.

2.1 Experimental

All the compounds have been isolated using the International Humic Substances Society (IHSS) protocol with some needful modifications.

2.2 Isolation of Land Fill Delhi (LFD) Compounds, LFD-I &LFD-II

1 g of LFD Inert soil was taken in 250 ml of the conical flask and 150 ml of 6N HCl was poured into it which was left overnight with stirring. This was filtered and the insoluble part was treated with 10% NaOH solution, left overnight, and again filtered. The aqueous portion was acidified with conc. HCl when 428 mg of the product LFD-I was collected by filtration. This filtrate was used in the next step. The above filtrate was left for complete drying in sunlight and the residue was extracted with methanol to get 312 mg of the compound LFD-II.

2.3 Isolation of LFD-III & LFD-IV

1 g of LFD Inert soil was taken in 250 ml of a conical flask and 150 ml of 6N HCl was poured into it and left overnight with stirring and filtered. The aqueous solution was neutralized with 10% NaOH and 632 mg of LFD-III was collected by filtration. The above aqueous filtrate was neutralized with 10% NaOH in excess and 726 mg of LFD-IV was obtained.

3. Results

3.1 UV-Visible Spectral Studies

UV-Visible Spectrum of LFD-I shows peaks at 209, 219, 239, 249, 284, and 364 nm. The UV-visible spectrum shows the absence of any strong aromatic chromophores. In the UV-Visible Spectrum of LFD-II, peaks were observed at 360 nm. The UV-Vis spectrum shows the absence of any strong aromatic chromophores. In the UV-Visible spectrum of LFD-III peaks were observed at 209, 211 nm. The above UV-Vis spectrum lacks any strong aromatic chromophores [21].

3.2 FT-IR Spectral Studies

FT-IR spectrum of LFD-I shows peaks at 1710.10, 2947.87, 3141.87, 3328.11 cm^{-1} along with other signals which show the presence of different functional groups like (COOH, OH, NH_2, and hydrogenated OH, etc.). FT-IR spectrum of LFD-II shows peaks at 1744.17, 2366.01, 2914.98, 3117.71, and 3318.40 cm^{-1} along with other signals which show the presence of different functional groups like (COOH, OH, NH_2, and hydrogenated OH, etc.). FT-IR spectrum of LFD-III shows peaks at 1667, 1701, 2923, 3117, and 3318 cm^{-1} along with other signals which show the presence of different functional groups like (COOH, OH, NH_2, and hydrogenated OH, etc.). FT-IR spectrum of LFD-IV shows peaks at 1527, 1813, 2374, 2862, 2854, 2929, and 3393 cm^{-1} along with other signals which show the presence of different functional groups like (COOH, OH, NH_2, and hydrogenated OH, etc.) [22].

3.3 NMR Spectral Studies

^{1}H-NMR spectrum of LFD-I showed signals at δ 1.190, and three signals at δ 3.319, 3.257, 3.516, and 8.013 ppm. No aromatic protons were observed [23].^{1}H-NMR Spectrum of LFD-II showed signals at δ 1.1872-1.9343, (m); 2.1929, 2.2229, (m); 3.2570-3.7449, (m) and 8.013 ppm. The value at δ 2.1 shows the presence of a proton of the cyclopropane ring.

3.4 Mass Spectral Studies of LFD Series

The mass spectrum of LFD-I is shown in (Figure 5).

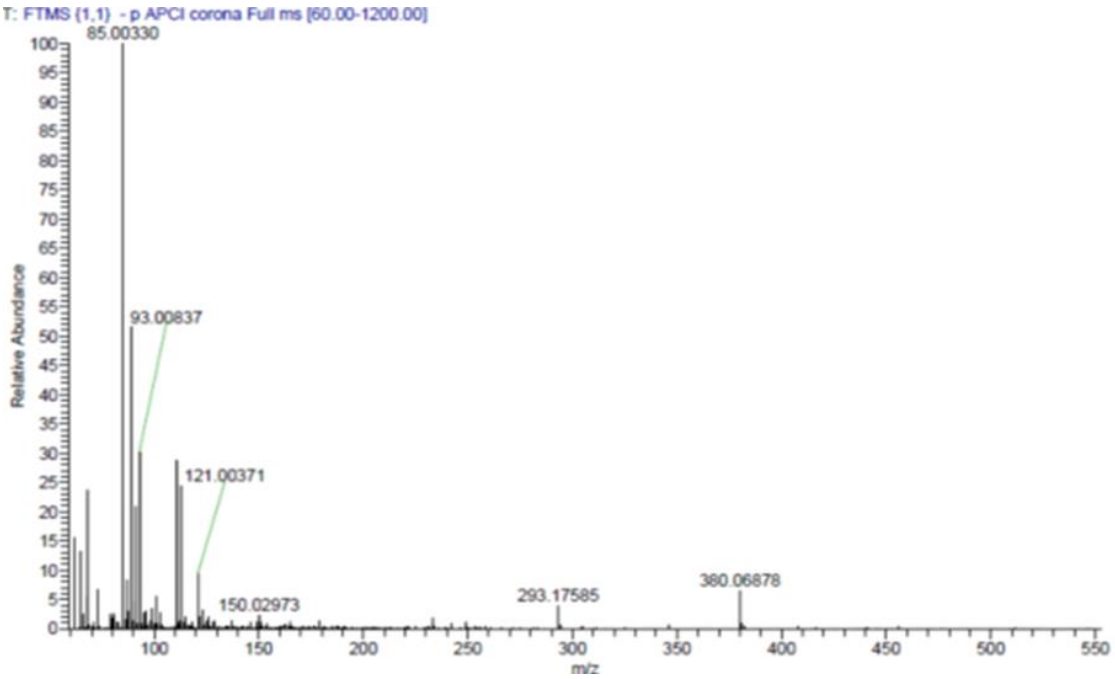

Figure 5 Mass spectrum of LFD-I.

The highest peak observed in the spectrum is at m/z 380.06476, representing the molecular ion (M^+-1H) and its Molecular weight is 381.06476 amu and the molecular formula is thus $C_{15}H_{11}NO_{11}$. Other fragments observed in the mass spectrum are at m/z 293.17585, 150. 00973, 121.00371 & 85. 00330. The loss of 231.01227 amu forms the observed peak at m/z at 150.00973. [381.06476 - 231.01227 = 150.05249].

The mass spectrum of LFD-II is shown (Figure 6).

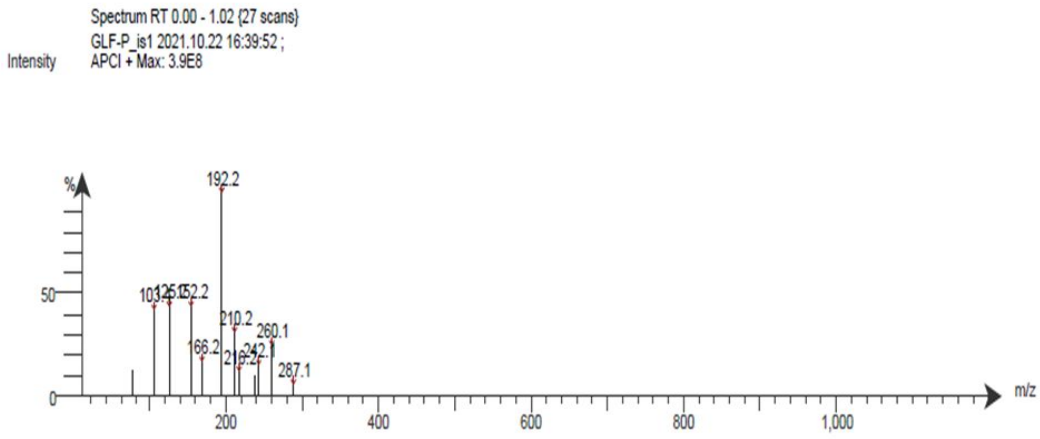

Figure 6 Mass spectrum of LFD-II.

In the Mass Spectrum of LFD-II (Positive ion mode), the peak observed was at m/z 287.1 (M^+-1H) along with other fragment peaks. The molecular weight of LFD-II is thus 288.108 amu and its molecular formula is $C_{11}H_{12}O_9$.

The mass spectrum of LFD-III is shown (Figure 7).

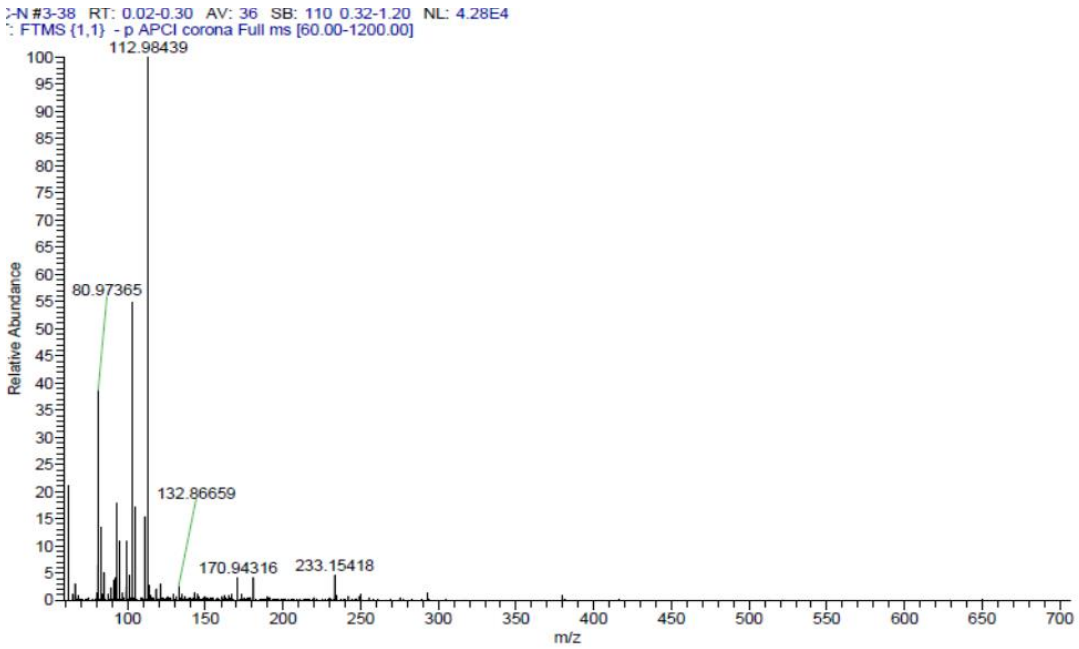

Figure 7 Mass spectrum of LFD-III.

In the Mass spectrum of LFD-III, the highest peak observed at m/z is 233.15418 amu along with other fragment peaks in the positive ion mode mass spectrum result of LFD-III. The molecular weight of LFD-III is thus 233.1263 amu and its molecular formula is $C_9H_{15}NO_6$. Other fragments observed are at m/z 170.95398, 132. 56657 & at 112. 98433 amu.

The mass spectrum of LFD-IV is shown (Figure 8).

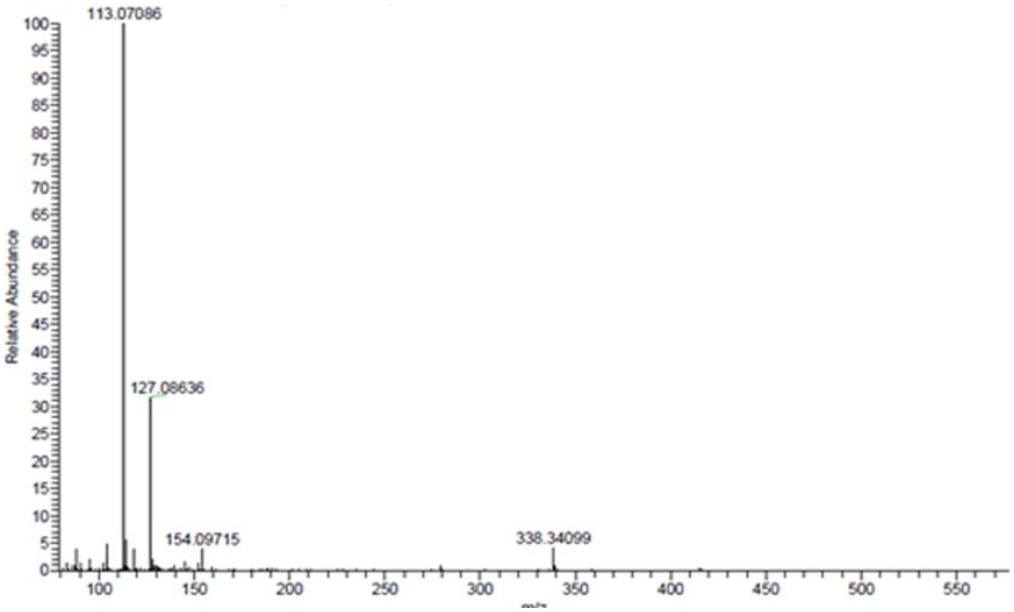

Figure 8 Mass spectrum of LFD-IV.

In the Mass spectrum of LFD-IV, the highest peak observed was at m/z 338.34099 amu along with other fragment peaks in the positive ion mode mass spectrum result of LFD-IV. The molecular weight of LFD-IV is 338.34099 amu and its molecular formula is $C_{14}H_{10}O_{10}$. Other fragments are observed at m/ z 154.09715, 127.08536, 113. 07086 amu.

From the above data, it can be surmised that LFD-I can be considered as the parent compound isolated from the Ghazipur landfill inert soil. The peak observed at 380.06476 is the (M^+- 1H) and hence the molecular weight of LFD-I is m/z 381.06476 amu with molecular formula $C_{15}H_{11}NO_{11}$ (error %-0.009). The major peak at m/z 150.02973 represents a loss of m/z 230 amu. The observed peak at m/z 150.02973 is (149.0448 + 1H = 150.02973). Addition of these values of m/z (150.02973 + 231.01227 = 381.042) (Calc.) which is the molecular weight of LFD-I.

The presence of the cyclopropane ring makes these molecules resemble Pyrethrins, which are isolated from the Chrysanthemum flowers. Synthetic Pyrethroids like Deltamethrin, Cypermethrin, and Allethrin are great commercial successes. The latter is used in common household mosquito mats. Similar insecticidal activity can be foreseen in the above compounds [24, 25].

3.5 SEM Studies

SEM studies on LFD-III and LFD-IV shown in (Figure 9), reveal the polycrystalline structure with the formation of slender fibers and smaller aggregates [26].

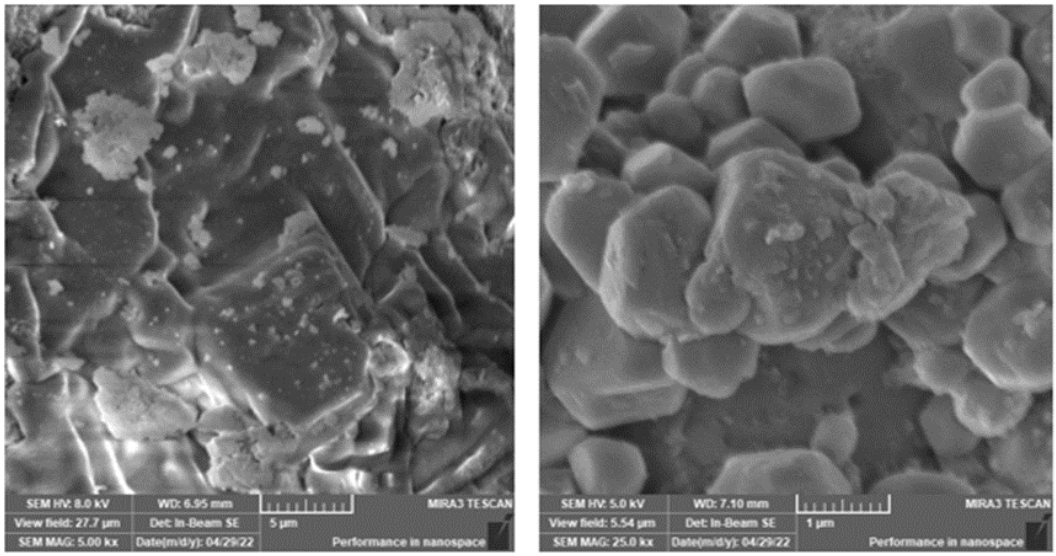

Figure 9 SEM images of (a) LFD-III at 5 µm (b) LFD-IV at 1 µm.

3.6 Computational Studies

Computational studies of these four compounds predicted their anti-bacterial, and anti-inflammatory activities using Instant. Chem. J. 21.15.0 software which gave the IC_{50} of 8.720, 8.42, and 6.99 (Table 2), which is a reasonably good value [27]. The new compounds are shown to be 5-HT2A, 5-HT2C, and H1 in L929 receptor-binding agents and CHO cells. These receptors are involved in brain diseases like Alzheimer's, Dementia, and Parkinson's. The CHO cells point towards their possible anti-cancer activity.

Table 2 Prediction of Biological Activity with IC_{50}.

Mol. Weight	LogP/MW	Rings	Atoms	Types	Value	Value. MIN	Value. MAX	Target Name	BIO.CELL
381.25	76.56	4	42	IC50	8.7200	8.72	8.72	5-HT2A	L929 Cell
381.25	76.56	4	42	IC50	8.4200	8.42	8.42	5-HT2C	CHO Cell
381.25	76.56	4	42	IC50	6.9900	6.99	6.99	H1	CHO Cell

Detailed Mass-spectral investigations, especially MS/MS studies and 2D-NMR spectral studies are in progress and will be published subsequently.

3.7 Recommendations

Strict regulations are necessary for the collection and transport of MSWs to appropriate budget allotments for building modern landfills must be, made both in the central and state budgets each year according to their needs.

The parliament and state legislatures supported by municipal corporations must pass the required laws and regulations with clauses for their strict implementation.

The current scenario of the MSW treatment plant in India has been given in (Table 3) below.

Table 3 Facilities for MSWM in major Indian states (Ministry of New and Renewable Energy, 2016).

S. No.	States	No. of units of treatment				
		Composting	Vermicompostin g	Biomethenation	Pelletization	Waste to Energy
1	Andhra Pradesh &Telangana	24	0	0	11	2
2	Andaman& Nicobar	1	0	0	0	0
3	Chandigarh	0	0	0	1	0
4	Delhi	3	0	0	0	3
5	Goa	14	0	0	0	0
6	Gujrat	3	93	0	6	0
7	Himachal Pradesh	10	0	0	0	0
8	Karnataka	0	0	0	0	0
9	Kerala	21	7	10	1	1
10	Madhya Pradesh	7	0	0	2	0
11	Maharashtra	6	2	5	5	2
12	Orissa	1	0	0	0	0
13	Punjab	1	3	0	0	0
14	Rajasthan	0	0	0	0	0
15	Tamil Nadu	102	24	0	3	0
16	Uttarakhand	0	0	0	0	0
17	Uttar Pradesh	0	0	0	0	0
18	West Bengal	13	7	0	0	0

A recent paper highlights the use of integrated MSW management methods. This paper also describes the current situation in major Indian cities. It is pointed out that Tamil Nadu leads the other states having 102 treatment plants whereas most others are in single or early double digits. It is suggested that such MSW treatment plants should be established in all States, Gram Panchayats, Blocks, and Districts.

3.8 Revenue Generation from the Landfills

Another author discusses the hierarchy in the management of Chandigarh city solid waste and emphasizes the importance of norms for field staff. The efforts of the Chandigarh Safai Karamchari Union, which has 3,450 members, have resulted in a 10% increase in their wages. Measures to ensure good health include using hand gloves, masks, soaps, anti-fungal, and hepatitis B vaccination, and social security measures.

In addition, strict implementation of Fines/challans for those who violate the law of the land in this regard could also be an additional source of income. Between 2011 and 2014, fines imposed in Chandigarh, Punjab State are 3,000-4,000 US dollars annually. In particular, collecting greenhouse

gases while protecting the environment will lead to generating additional resources. Even a solid waste management tax has also been suggested [28].

4. Discussion

Landfills emit harmful gases (CO_2, CH_4, CO, HCHO, H_2S, and NH_3) and occasional fires. An example of a recent study is reported from Gorakhpur, Uttar Pradesh, India. This study compiles the emission of gases and toxic chemicals from landfills in that area [29].

We have isolated new compounds from the landfill-inert soil, Ghazipur using the International Humic Substances Society (IHSS) method with some modifications and characterized these various techniques. The UV-visible spectrum of these compounds shows the absence of any aromatic or strongly absorbing chromophore, and FT-IR spectra show the presence of many functional groups such as COOH, OH, NH_2, etc. NMR studies indicate the presence of cyclopropane ring. These compounds' molecular weights and formulae have been determined using high-resolution mass spectrometry. SEM studies show the crystalline nature of the compounds, whereas computational studies show these compounds can bind to the 5HT2A and 5HT2C receptors. These receptors are known to be involved in neurodegenerative diseases like Parkinson's, Alzheimer's, and Dementia.

5. Conclusions

Four new compounds have been isolated from the inert soil obtained from the Ghazipur Municipal Solid waste. Their structures resemble the natural Pyrethrins (originally identified in Chrysanthemum flowers), serving as well-known environmentally safe insecticides. Thus looking for value-added products from composted city solid waste is attractive not only for adding to the economy but also, in the long run, these could lead to environmental cleaning and contain the adverse effects of climate change [30]. This entire field of study must be considered a highly interdisciplinary area of study; hence, experts from different fields must be involved in finding the long-terms solution.

Abbreviations

IHSS	International Humic Substances Society
LFD	Landfill Delhi
MSW	Municipal Solid Waste
EDMC	East Delhi Municipal Corporation
RCRA	The Resource Conservation and Recovery
EPA	Environmental Protection Agency
UV spectroscopy	Ultraviolet-visible Spectroscopy
FT-IR	Fourier Transform Infrared Spectroscopy
NMR	Nuclear Magnetic Resonance
SEM	Scanning Electron Microscopy
HRMS	High Resolution Mass Spectrometry

µM	Micro meter

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Author Contributions

SKT-Experimental and computational work, SVE- Conceptualization, planning and drafting of the manuscript.

Competing Interests

The authors declare no conflict of interest.

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